

Reactive thrombocytosis in pulmonary tuberculosis

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SUMMARY The incidence of reactive thrombocytosis in active pulmonary tuberculosis was studied in 122 patients. Thrombocytosis was common, platelet counts often exceeding $1 \times 10^{12}/l$. A significant inverse correlation was noted between the mean platelet volume and the platelet count ($r = -0.54$, $p < 0.0001$). Interval estimation suggested that this relation was non-linear. Further studies were done in a small group of six patients. Platelet survival was considerably shortened, the platelets aggregated excessively in vitro, serum concentrations of thrombopoiesis stimulating activity were raised, and serotonin uptake and release were within normal limits. The degree of thrombocytosis correlated significantly with the degree of inflammation measured by the erythrocyte sedimentation rate ($r = 0.40$, $p < 0.003$) and serum C-reactive protein concentration ($r = 0.35$, $p < 0.008$).

Thrombocytosis occurs in many chronic inflammatory diseases, including tuberculosis.¹⁻⁴ The precise stimulus for increased platelet production in reactive thrombocytosis is not clear, but it is associated with increased numbers of small megakaryocytes in the marrow,⁴⁻⁶ which show reduced nuclear ploidy.⁵ The platelets are also small,⁷⁻⁹ but it has recently been suggested, that this may simply reflect the thrombocytosis, as there is normally an inverse correlation between the number and volume of platelets.¹⁰ The present study was done to define reactive thrombocytosis in acute tuberculosis with particular reference to the association between the volume and number of platelets, their survival, and certain aspects of their function.

Patients and methods

One hundred and twenty two patients with pulmonary tuberculosis were studied; most were of black or mixed racial origin. Twenty were newly diagnosed and about to begin antituberculous treatment, 82 had been diagnosed recently and were already being treated, and 20 were nearing the end of a six month course of treatment. A full blood count was done in

all patients. Acute phase markers, including the erythrocyte sedimentation rate (ESR) and serum C-reactive protein (CRP) concentration were measured in 58 subjects, and thrombopoiesis stimulating activity was assessed in six untreated controls. Platelet function tests and platelet survival were also measured in five of these six subjects. Platelet counts were done weekly in a further seven patients to assess the effects of treatment on the reactive thrombocytosis.

Full blood counts, including platelet counts and platelet indices, were performed on a Coulter Model-S-Plus electronic counter (Coulter Electronics, Hialeah, Florida, USA) with standard calibration and quality control. CRP concentrations were measured by an immunoturbidometric method¹¹ on an IL III multistat centrifugal analyser (Instrumentation Laboratory, Lexington, Massachusetts, USA). CRP standards were obtained from Behringwerke (Marburg, West Germany) and antibodies to CRP from Dako (Copenhagen, Denmark). Serum thrombopoiesis stimulating activity was measured in a bioassay system¹² and the results were compared with those from three normal subjects whose activity was assigned the value of 100%. In the platelet survival studies autologous platelets were labelled with ¹¹¹Indium oxine and then reinfused. Survival was evaluated by the method of Thakur *et al*,¹³ as modified by Heyns *et al*.¹⁴ Platelet aggregation was

measured by the method of Born and Cross¹⁵ using a Chronolog Dual Channel Aggregometer (Coulter Electronics, Hialeah, Florida, USA) attached to a Coulter Electronics Omniscribe recorder (Coulter Electronics, Hialeah, Florida, USA). Platelet uptake and release of radioactive serotonin were measured by the method of David and Herion.¹⁶

The study was approved by the Committee for Research in Human Subjects of the University of the Witwatersrand. Subjects were admitted to the study only after it had been explained to them and their informed consent obtained.

Linear correlation was determined by the Pearson product-moment coefficient of correlation. Confidence limits and residuals of the appropriate regression line were calculated by regression analysis using the statistical analysis system on an IBM 370 computer. Differences between discrete platelet count intervals in terms of mean platelet volumes were determined both by analysis of variance and by sequential Student's *t* tests with a Bonferroni correction.

Results

The mean platelet count in the 20 patients about to start treatment was $582.6 \times 10^9/l$ (SD 194.8), $681.0 \times 10^9/l$ (SD 232.0) in the 82 who had just started treatment, and $292.4 \times 10^9/l$ (SD 75.9) in the 20 nearing the end of the course. A normal range of $159\text{--}415 \times 10^9/l$ (mean $283 \times 10^9/l$) was established in 564 normal subjects of black, white, and mixed race. There were no statistical differences between

platelet counts in the first two groups but both were significantly different from the group nearing the end of the course ($t = 6.2$, $p < 0.0005$ and $t = 7.4$, $p < 0.0005$, respectively). Seven patients who were already being treated, and who had reactive thrombocytosis, had platelet counts weekly for 11 weeks. There was a steady, though modest, drop from the seventh week onwards, the final mean value being about 70% of the original.

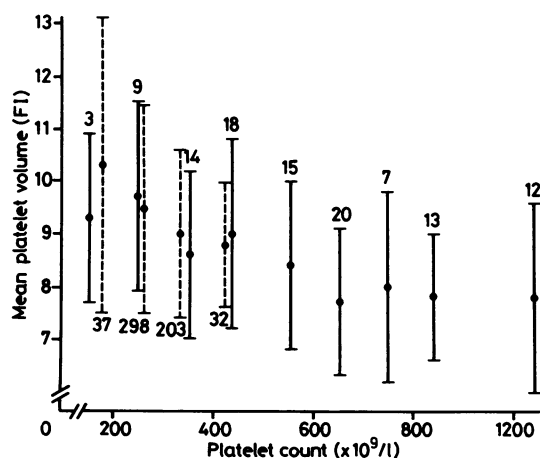
The mean platelet volume was normally distributed, with a skewness of 0.3941. There was a significant inverse correlation between mean platelet volume and platelet number, with a Pearson product-moment coefficient of correlation of -0.54 ($p < 0.0001$). Interval analysis suggested that the inverse correlation was not linear (figure). Platelet volumes in the lower ranges were significantly different from those in the higher ones. The fact that there was no significant difference between mean platelet volumes for intervals at higher platelet counts emphasised that the volume number inverse relation was not linear. In a further analysis the values of mean platelet volume for the various intervals in the patients with tuberculosis were compared with those in the 564 normal subjects. There was no significant difference between the two groups at any interval over the normal range (figure).

In vitro platelet aggregation in five of the subjects suggested that their platelets might aggregate abnormally; this was best illustrated by the aggregation in response to arachidonic acid. The minimal doses inducing full aggregation were 0.4, 0.2, 0.3, 0.3 and 0.3 mmol (normal minimal dose 0.5–1.0 mmol). In contrast, serotonin uptake and release remained normal. Platelet survival was reduced and concentrations of thrombopoiesis stimulating factor were significantly raised in the five subjects (table).

The relation between the increased platelet count and the activity of the disease was assessed further in 58 patients receiving treatment. There was a significant direct correlation between the ESR and the platelet count ($r = 0.40$, $p < 0.003$) and between the serum CRP concentration and the platelet count ($r = 0.35$, $p < 0.008$). The CRP and ESR correlated significantly with each other ($r = 0.50$; $p < 0.0001$).

Discussion

The association between tuberculosis and thrombocytosis has been reported^{1,2} but as an uncommon occurrence,¹⁷ and in a 1974 review of the haematological complications of tuberculosis it was not even mentioned.¹⁸ Though our study was not designed to assess prevalence, it seemed that reactive thrombocytosis was common in our group of patients with active pulmonary tuberculosis, and that it could



Non-linear correlation between mean platelet volume (2 SD) and given platelet intervals in normal subjects (----) and patients (—), with reactive thrombocytosis in pulmonary tuberculosis.

Table Data on platelet survival, serum thrombopoiesis stimulating activity, and platelet counts in six patients with recently diagnosed active pulmonary tuberculosis

Measurement	Case 1	Case 2	Case 3	Case 4	Case 5	Case 6	Normal values
Mean platelet survival (days)	2.0	2.5	2.5	1.4		3.0	7-11
Thrombopoiesis stimulating activity (%)*	142†	122†	120†	140†	115†	120†	100
Platelet count ($\times 10^9/l$)	835	708	830	728	460	829	160-415

*Relative to the mean thrombopoiesis stimulating activity in three normal subjects.

†Significantly greater than controls ($p < 0.05$).

be severe, with eight (9%) of the 92 having platelet counts of $> 1000 \times 10^9/l$.

The present findings have shown an inverse, non-linear relation between mean platelet volume and platelet count in active tuberculosis, and also that platelet survival is considerably shortened. The correlation between high platelet counts and decreased platelet life span shows that thrombopoiesis must be increased. The finding that many of the circulating platelets are both young and small in reactive thrombocytosis runs counter to the hypothesis that young platelets tend to be large.^{19,20} Why they should be small is not known, although it may be associated with the fact that the megakaryocytes in states of reactive thrombocytosis tend to be small.⁴⁻⁶

The possibility that the increased numbers of small young platelets might be a feature of pulmonary tuberculosis was also considered. The pulmonary vasculature is thought to contribute to the production of platelets by fragmenting proplatelets,²¹ and the question arose as to whether a diseased microvasculature in pulmonary tuberculosis might lead to excessive fragmentation and hence smaller platelets. Such an explanation seems untenable, however, as a parallel study on the reactive thrombocytosis of rheumatoid arthritis gave results identical with those in pulmonary tuberculosis (unpublished observations).

The raised concentrations of serum thrombopoiesis stimulating activity in patients with pulmonary tuberculosis suggest the presence of a circulating stimulatory factor, which is in keeping with a previous report of increased thrombopoietin concentrations in patients with tuberculosis.²² It must be emphasised, however, that the bioassay used in the present study cannot distinguish between diverse stimulators of different chemical composition. The fact that small platelets are produced in reactive thrombocytosis suggests that the inflammatory mediator is different from the one operating in idiopathic thrombocytopenia, because the platelets are large in this condition. Therefore, the large megakaryocytes of high nuclear ploidy, which are a feature of idiopathic thrombocytopenia,⁵ may be the result of stimulation via a mediator induced by thrombocytopenia, and the

small megakaryocytes of low nuclear ploidy, which occur in reactive thrombocytosis,⁵ may result from an inflammation induced stimulator.

Our finding that platelets can aggregate excessively in reactive thrombocytosis in tuberculosis is at variance with the observations of Thompson *et al.*²³ It is, however, compatible with a recent report of synergism between platelet agonists and the acute phase reactant, CRP.²⁴ In the 58 patients in the present study in whom CRP was evaluated the mean serum concentration was 60.8 (SD 37.2) mg/l (normal values < 12 mg/l). Therefore, the increased quantities of CRP, which were present in the platelet rich plasma, might have been acting in concert with aggregation agonists to produce excessive aggregation.

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